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Copper complexes with sulfonamides: crystal structure and interaction with pUC18 plasmid and hydrogen peroxide

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Abstract

N-Quinolin-8-yl-benzenesulfonamide (Hqbsa) and *N*-quinolin-8-yl-naftalenesulfonamide (Hqnsa) have been synthetized and physicochemically characterized, and used as ligands to coordinate copper complexes with ML2 stoichiometry. The structure of the compounds $[Cu(qbsa)_2]\cdot DMF$ and $[Cu(qnsa)_2]$ has been determined by X-ray diffraction and both of them crystallize in the orthorhombic system. IR and ESR spectra of the complexes are discussed. The cleavage of pUCI8 by the copper complexes do not behave as chemical nucleases in the range of concentrations assayed.

Keywords: Copper(II) complexes; Sulfonamides; Crystal structures; DNA interaction

1. Introduction

Many studies have dealt with the application of transition metal complexes in probing nucleic acids and the recognition of specific sites on the nucleic acid polymer [1–4]. Within this field, one of the widest studied subject has been the coordination chemistry of copper with ligands containing *N*-donor atoms belonging to aromatic heterocycles, mainly after it was demonstrated that copper bis-phenantroline complexes are able to catalyze the cleavage of nucleic acids in the presence of H_2O_2 and a reducing agent [5–8]. It has been previously shown [9] that DNA does not interact significantly with a copper complex containing *N*-quinolin-8-yl-*p*-toluensulfonamide. As the de presence of the organic rings close to the cooper site plays an important role in binding the complex to de DNA chain, two new complexes of cooper with ligands similar to that previously used have been synthetized and characterized, and their interaction with pUCI8 plasmid has been studied.

2. Experimental

2.1. Materials and methods

8-Aminoquinoline and sulfonyl chlorides were provided by Fluka AG and all reagents used were of analytical grade.

Chemical analyses for carbon, hydrogen, and nitrogen were performed on a 2400 elemental analyser from Perkin–Elmer. Copper was determined on a ICP spectrometer (Perkin–Elmer model 2380 Plasma 2).

FT IR spectra were recorded using KBr mulls and a Perkin–Elmer FT-IR 1730 instrument. Electron paramagnetic resonance spectra were recorded at X-band frequencies with a Bruker ER 200D. Molecular masses were measured by Servicio de Masas (Universidad Autónoma de Madrid, Spain) by de FAB method with samples held on a nitrobenzyl alcohol (NBA) matrix and L-SIMS ionisation mode, in a VG AUTOSPEC.

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2.2. Synthesis of the compounds

The ligands *N*-quinolin-8-yl-benzenesulfonamide (Hqbsa) and *N*-quinolin-8-yl-naftalene-2-sulfonamide (Hqnsa), were prepared by reaction of 8-aminoquinoline with the corresponding sulfonyl chlorides, following the meted described elsewhere [9]. Elemental chemical analysis data are in agreement with the formulae $C_{15}H_{12}N_2O_2S$ and $C_{19}H_{14}N_2O_2S$ for Hqbsa and Hqnsa, respectively.

The copper complexes were prepared by direct synthesis between Hqbsa and Hqnsa with soluble copper salts, as previously described [9] and the elemental chemical analysis data are in agreement with the formulae $CuC_{33}H_{29}N_5O_5S_2$ and $CuC_{38}H_{26}N_4O_4S_2$, respectively.

2.3. X-ray determination structure

A blue needle crystal of $[Cu(qbsa)_2]\cdot DMF$ and a red prismatic crystal of $[Cu(qnsa)_2]$ were mounted on a glass fibber and used for data collection. For the $[Cu(qbsa)_2]\cdot DMF$ complex, cell constants and an orientation matrix for data collection were obtained by least-squares refinement of the diffraction data on an Enraf Nonius CAD4 automatic diffractometer [10]. Data were collected at 293 K using Cu Kα radiation ($\lambda = 1.54184$ Å) and the ω-scan technique, and corrected for Lorentz and polarization effects [11]. A semiempirical absorption correction (Ψ-scan) was made [12].

For the $[Cu(qnsa)_2]$ complex, crystal data were collected at 291 K using a Bruker SMART CCD 1000 diffractometer. Graphite monochromated Mo Kα radiation ($\lambda = 0.71073$ Å) was used throughout. The data were processed with SAINT [13] and corrected for absorption using SADABS (transmissions factors: 1.000-0-518) [14].

Both structures were solved by direct (Patterson and Fourier) methods [15], and subsequent difference Fourier maps, which revealed the position of all non-hydrogen atoms, and refined on F^2 by a full-matrix least-squares procedure using anisotropic displacement parameters [16]. Hydrogen atoms were placed geometrically and positional parameters were refined using a riding model. Atomic scattering factors were taken from 'International Tables for X-ray Crystallography' [17] and molecular graphics from PLATON [18]. A summary of the crystal data, experimental details and refinement results are listed in Table 1.

Table 1
Summary of crystal parameters, data collection and refinement for the two crystal structures

	$[Cu(qbsa)_2]\cdot DMF$	$[Cu(qnsa)_2]$
Empirical formula	$CuC_{33}H_{29}N_5O_5S_2$	$CuC_{38}H_{26}N_4O_4S_2$
Formula weight	703.27	730.29
Temperature (K)	213(2)	293(2)
Wavelength (Å)	1.54184	0.71073
Crystal system	orthorhombic	orthorhombic
Space group	*Pbca* (No. 61)	*P*4(1) (No. 74)
Unit cell dimensions		
a (Å)	17.008(3)	17.879(2)
b (Å)	17.365(16)	17.879(2)
c (Å)	21.651(4)	21.511(3)
α (°)	90	90
β (°)	90	90
γ (°)	90	90
Volume (Å^3)	6394.6(17)	6876.2(16)
Z	8	8
Calculated density (g cm^{-3})	1.461	1.411
Absorption coefficient (mm^{-1})	2.600	0.803
F (000)	2904	3000
Crystal size (mm)	0.35 × 0.10 × 0.05	0.39 × 0.15 × 0.15
θ Range for data collection (°)	5.09–65.11	1.48–28.07
Index ranges	$-1 < h < 20$, $-1 < k < 17$, $-1 < l < 25$	$-20 < h < 23$, $-23 < k < 23$, $-28 < l < 21$
Reflections collected, unique, R_{int}	6351, 5264, 0.0827	35 424, 12 759, 0.1317
Completeness to $\theta = 65.11$ (%)	87.6	97.2
Absorption correction	'Psi-scans'	SADABS
Max/min transmission	0.8810 and 0.4631	0.8890 and 0.7446
Refined method	full-matrix least-squares on F^2	full-matrix least-squares on F^2
Data, restraints, parameters	5264/0/415	12759/1/884
Goodness-of-fit on F^2	0.993	0.793
Final *R* indices $[I > 2\sigma(I)]$	$R_1 = 0.0704$, $wR_2 = 0.1341$	$R_1 = 0.0807$, $wR_2 = 0.1445$
R indices (all data)	$R_1 = 0.2037$, $wR_2 = 0.1765$	$R_1 = 0.2569$, $wR_2 = 0.1952$
Largest difference peak and hole (e Å^{-3})	0.578 and −0.453	0.339 and −0.601

2.4. Cleavage of pUCI8 by the copper complex

The copper complexes are insoluble in the different solvents used in cleavage processes such as Tris–HCl; dihydrogenphosphate–hydrogenphosphate etc.; consequently we must use a solution in DMF following the experimental conditions reported by Reddy [19].

A typical reaction was carried out by mixing 3 μl of the Cu(II) complex (100 μM in DMF), 2 μl of 0.25 μg μl^{-1} pUCI8 and 3 μl of H_2O_2 10 mM. The resulting solution contains 15 μM of the complex, 0.025 μg μl^{-1} pUCI8 and 1.5 mM of H_2O_2. After allowing the sample to incubate at 25 °C for 3 h, 3 μl of a quench buffer solution consisting of 0.25% bromophenol blue, 0.25% xylene cyanole and 30% glycerol was added. Then the solution was subjected to electrophoresis on a 0.7% agarose gel in 1 × TBE buffer (0.045 M Tris, 0.045 M

boric acid and 1 mM edta) at 80 V about 2 h. The gel was stained with 10 μg ml^{-1} ethidium bromide and photographed on a capturing system gel printer plus TDI.

3. Results and discussion

The structures of the two sulfonamides synthetized are shown in Scheme 1.

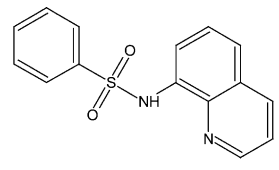
Reaction of these sulfonamides with Cu(II) salts deprotonates the N(sulfonamid) atom, thus coordinating through both N atoms to yield complexes with a ML_2 stoichiometry, that is, $[Cu(qnsa)_2]$, but $[Cu(qbsa)_2]\cdot DMF$, as this last complex crystallizes with a molecule of DMF (from the solvent).

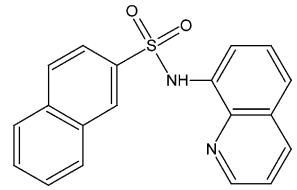
These formulae are also in agreement with mass spectrum data. So, complex $[Cu(qbsa)_2]\cdot DMF$ gives rise to a signal at $m/z = 629.9$, which coincides with that of the molecular ion once the DMF molecule has been removed, probably weakly held to the Cu(II) complex. Loss of one of the ligands is in agreement with a peak at $m/z = 347$ also recorded. Complex $[Cu(qnsa)_2]$ gives rise to two important peaks at $m/z = 730$ and 396, corresponding to the molecular ion and to a $[Cu(qnsa)]^+$ fragment, respectively.

3.1. Description of the crystal structures

The crystalline structure of the complex $[Cu(qbsa)_2]\cdot DMF$ is shown in Fig. 1 (the DMF molecule is not included for clarity). Complex $[Cu(qnsa)_2]$ is obtained as a racemic mixture with two units in the asymmetric unit (molecules a and b, see Fig. 2). Significant bond distances and angles are summarized in Table 2.

Sulfonamide acts as a bidentate ligand, forming five-member rings. The Cu–N distances are similar to those found in similar Cu complexes [9,20,21], although Cu–N(sulfonamide) are slightly shorter than the Cu–N(quinoline) distances, 1.884–1.950 versus 1.973–2.000 Å. The Cu–O distances are close to 3 Å, indicating





Scheme 1. Structure of the Hqbsa (top) and Hqnsa (bottom).

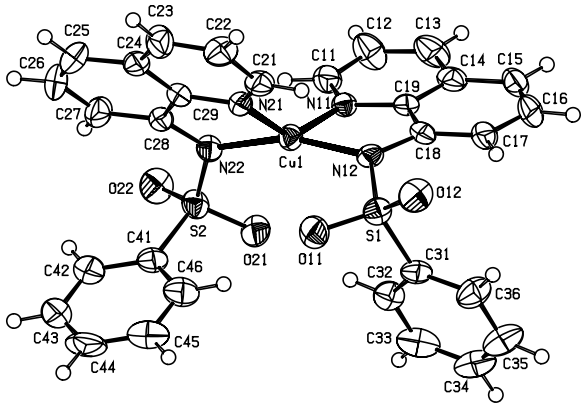


Fig. 1. ORTEP diagram for the $[Cu(qbsa)_2]\cdot DMF$ complex with the atom-labelling scheme.

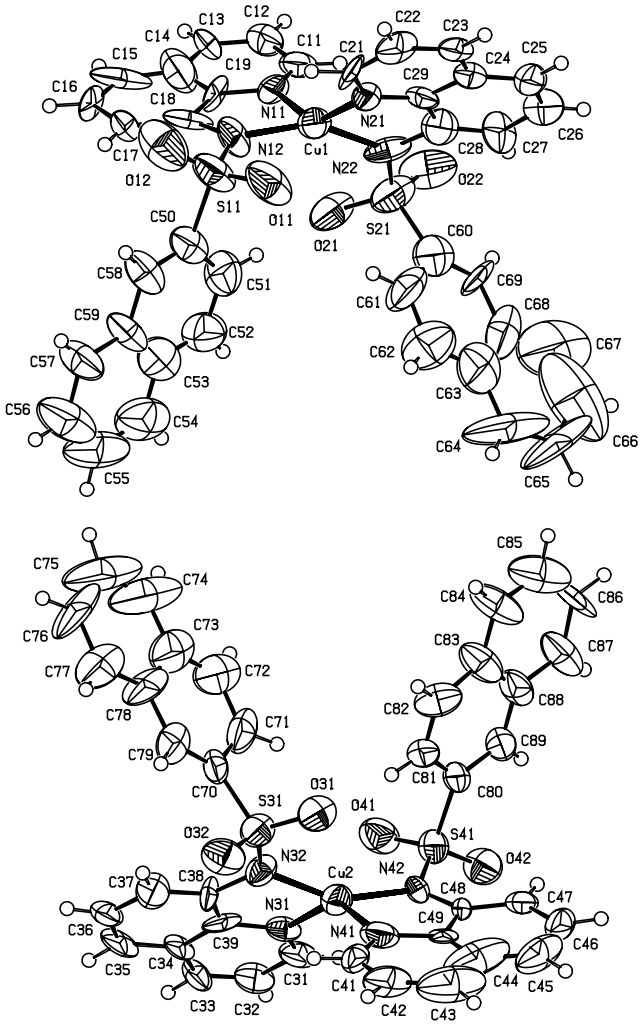


Fig. 2. ORTEP diagram for the $[Cu(qnsa)_2]$ complex (molecules a and b) with the atom-labelling scheme.

the lack of bonding between copper and oxygen atoms from the sulfonyl groups. Local environment around the Cu(II) cations is highly distorted. Tetrahedrality of the CuN_4 chromophore for the $[Cu(qbsa)_2]\cdot DMF$ complex

Table 2
Selected bond lengths (Å) and angles (°) for copper complexes

$[Cu(qbsa)_2]\cdot DMF$	
Bond lengths	
Cu(1)–N(12)	1.930(6)
Cu(1)–N(22)	1.932(6)
Cu(1)–N(11)	1.987(6)
Cu(1)–N(21)	1.993(6)
S(1)–N(12)	1.588(6)
S(2)–N(22)	1.581(6)
Bond angles	
N(12)–Cu(1)–N(22)	161.2(3)
N(12)–Cu(1)–N(11)	82.9(3)
N(22)–Cu(1)–N(11)	104.5(3)
N(12)–Cu(1)–N(21)	102.8(3)
N(22)–Cu(1)–N(21)	83.3(2)
N(11)–Cu(1)–N(21)	138.3(2)
S(1)–N(12)–Cu(1)	121.2(3)
S(2)–N(22)–Cu(1)	121.8(3)
$[Cu(qnsa)_2]$ (*a*)	
Bond lengths	
Cu(1)–N(22)	1.884(12)
Cu(1)–N(12)	1.932(10)
Cu(1)–N(11)	1.973(10)
Cu(1)–N(21)	1.985(13)
S(11)–N(12)	1.583(11)
S(21)–N(22)	1.593(11)
Bond angles	
N(22)–Cu(1)–N(12)	152.3(5)
N(22)–Cu(1)–N(11)	102.4(5)
N(12)–Cu(1)–N(11)	83.7(5)
N(22)–Cu(1)–N(21)	83.0(5)
N(12)–Cu(1)–N(21)	110.0(5)
N(11)–Cu(1)–N(21)	139.5(5)
S(11)–N(12)–Cu(1)	127.5(7)
S(21)–N(22)–Cu(1)	123.9(8)
$[Cu(qnsa)_2]$ (*b*)	
Bond lengths	
Cu(2)–N(42)	1.927(8)
Cu(2)–N(32)	1.950(9)
Cu(2)–N(31)	1.978(12)
Cu(2)–N(41)	2.000(10)
S(31)–N(32)	1.582(10)
S(41)–N(42)	1.596(10)
Bond angles	
N(42)–Cu(2)–N(32)	153.3(4)
N(42)–Cu(2)–N(31)	81.4(5)
N(32)–Cu(2)–N(31)	105.4(5)
N(42)–Cu(2)–N(41)	110.4(5)
N(32)–Cu(2)–N(41)	82.6(4)
N(31)–Cu(2)–N(41)	137.0(4)
S(31)–N(32)–Cu(2)	122.7(6)
S(41)–N(42)–Cu(2)	122.6(6)

is 48.62 and for the $[Cu(qnsa)_2]$ complex is 52.54 (molecule a) and 55.07 (molecule b).

3.2. FT IR spectroscopy

The positions of relevant bands in the FT IR spectra of both ligands and complexes are given in Table 3. The bands do not shift excessively from the ligands to the complexes, but some differences are worthwhile to be mentioned. The band due to N–H stretching mode in the spectra of the ligands has vanished in the spectra of the complexes, confirming coordination takes place through deprotonated N atoms. Consequently vibrations involving bond close to these N atoms result also affected, and thus the band due to the S–N bond is shifted 40 cm^{-1} to higher wavenumbers in the complexes than in the ligands, while the S=O band shifts 40 cm^{-1} to lower wavenumbers. This shift cannot be related to a direct interaction with the Cu(II) ions, as concluded from the XRD data above described. Splitting of the ν(S=O) band in the spectra of the complexes is probably due to non-equivalence of both SO_2 in the molecule.

3.3. EPR spectroscopy

The EPR spectrum of the polycrystalline complex $[Cu(qbsa)_2]\cdot DMF$ is axial with a slightly observed hyperfine splitting; as a consequence we have doped the complex with Zn(II). Fig. 3 shows the EPR spectrum together with the simulated one with the WIMSINFONIA program [22]. The EPR parameters deduced from the simulation program are $g_{||} = 2.235$, $g_{\perp} = 2.055$ and $A_{||} = 80 \times 10^{-4}$ cm^{-1}. The value of the $g_{||}/A_{||}$ ratio is 279, that according to Addison et al. [23], indicates a distorted tetrahedral geometry, as we can observe from the crystal structure.

The EPR spectrum of the powder complex $[Cu(qnsa)_2]$ presents a similar pattern to that of the complex $[Cu(qbsa)_2]\cdot DMF$. The EPR parameters deduced from simulation in doped copper–zinc complex are $g_{||} = 2.205$, $g_{\perp} = 2.078$ and $A_{||} = 105 \times 10^{-4}$ cm^{-1}. The value of $g_{||}/A_{||}$ is 210, suggesting also a distorted tetrahedral geometry. These results are in agreement with the values of the tetrahedrality deduced from the crystal structures.

3.4. Cleavage of pUCI8 by copper(II) complexes and copper(II) salt

Fig. 4 shows the results of electrophoresis between the complexes and the Cu(II) salt in the conditions reported in the Section 2. From this we can appreciate that none of the complexes behaves as chemical nucleases in the range of concentrations assayed (100–500 μM). In contrast, the copper(II) salt studied breaks the DNA supercoiled to nicked and linear DNA forms in the same range of concentrations. As a consequence, in the conditions of the experiment the complexes are non toxic to DNA, contrary to the behavior of the free copper(II).

In the presence of oxygen donor species, copper is thought to be able to form different oxidative inter-

Table 3
Selected IR bands (cm^{-1})

		Hqbsa	Hqnsa	$[Cu(qbsa)_2]\cdot DMF$	$[Cu(qnsa)_2]$
	ν_{N-H}	3218	3245		
SO_2	ν_{as}	1359	1375	1322	1322
				1286	1298
	ν_s	1169	1164	1145	1145
				1115	1127
	δSO_2	585	558	599	569
		559	546	566	549
	ν_{S-N}	926	918	958	962
	ν_{C-S}	686	679	691	658

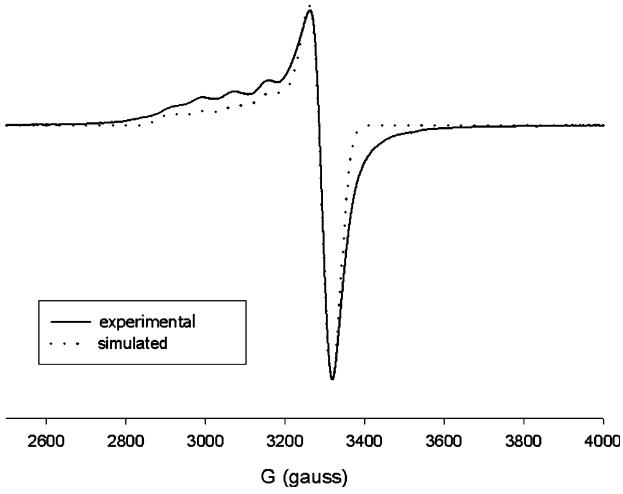


Fig. 3. EPR spectra for the $[Cu(qbsa)_2]\cdot DMF$ complex.

mediates, depending on the specific complex and conditions. A non diffusible copper–oxene intermediate has been invoked in some cleavage reactions [4,5,24] while, Fenton-type chemistry, which involves release of diffusible hydroxyl radical, has been suggested in other cases [25,26].

In all the proposed mechanisms, a redox reaction between copper(II) and the reducing agent must take place, producing an oxygen intermediate that cleaves the DNA. If the complexes reported here do not present chemical nuclease activity this is a consequence probably of their stability against the redox processes.

4. Supplementary material

Atomic coordinates and equivalent isotropic displacement parameters, bond lengths and angles, anisotropic

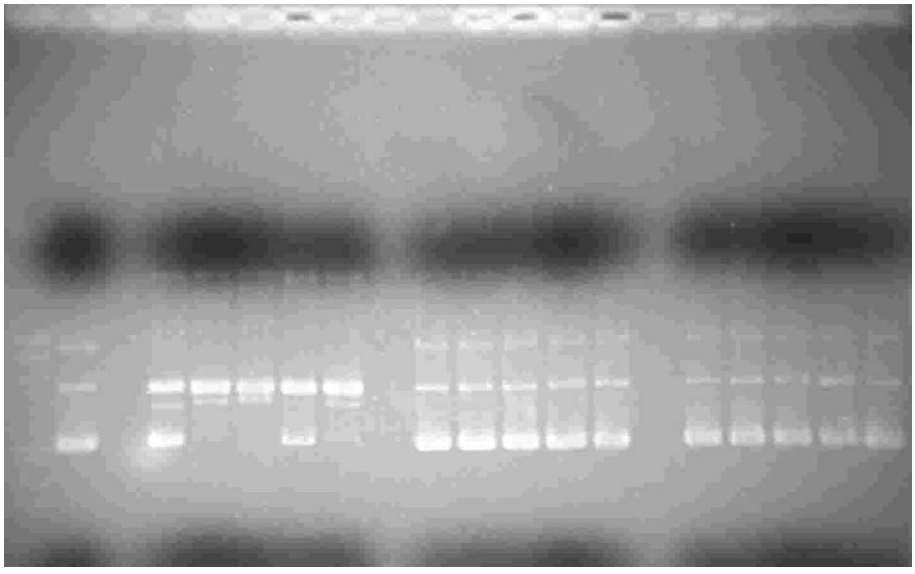


Fig. 4. Cleavage of pUCI8 by copper(II) complexes in the presence of H_2O_2. Lanes (1) Lambda DNA/ECORI+Hind III marker; (2) untreated pUCI8; (3) with added 100 μM Cu(II) and 2 mM H_2O; (4) with added 200 μM Cu(II) and 20 mM H_2O_2; (5) with added 300 μM Cu(II) and 30 mM H_2O_2; (6) 400 μM Cu(II) and 40 mM H_2O_2; (7) 500 μM Cu(II) and 50 mM H_2O_2; (8) 100 μM of the $[Cu(qbsa)_2]$ and 10 mM H_2O_2; (9) 200 μM $[Cu(qbsa)_2]$ and 20 mM H_2O_2; (10) 300 μM $[Cu(qbsa)_2]$ and 30 mM H_2O_2; (11) 400 μM of the $[Cu(qbsa)_2]$ and 40 mM H_2O_2. (12) 500 μM of the $[Cu(qbsa)_2]$ and 50 mM H_2O_2; (13) 100 μM of the $[Cu(qnsa)_2]$ and 10 mM H_2O_2; (14) 200 μM $[Cu(qnsa)_2]$ and 20 mM H_2O_2; (15) 300 μM $[Cu(qnsa)_2]$ and 30 mM H_2O_2; (16) 400 μM of the $[Cu(qnsa)_2]$ and 40 mM H_2O_2; (17) 500 μM of the $[Cu(qnsa)_2]$ and 50 mM H_2O_2.

displacement parameters, hydrogen coordinates and isotropic thermal parameters, hydrogen bonds and observed and calculated structure factors are available from the authors upon request.

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